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Chitosan-pDNA nanoparticle characteristics determine the transfection efficacy of gene delivery to human mesenchymal stem cells.

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Abstract

PURPOSE: This study evaluated the potential for prepared chitosan-plasmid DNA (pDNA) nanoparticles to transfer an exogenous gene into human bone marrow-derived mesenchymal stem cells (MSCs).

METHODS: Chitosan-pDNA nanoparticles were synthesized by the complex coacervation method. We used 18, 50 and 136 KD chitosan at concentrations of 0.05%, 0.1%, 0.5% and 1%, in addition to a pTracer-CMV2 plasmid that contained the green fluorescent protein (GFP) gene. To examine the complexation, samples were run through an agarose gel. The sizes and zeta potential of the nanoparticles were measured by a nanosizer. Scanning electron microscopy (SEM) imaging was used to observe the nanoparticle morphology. MSCs were prepared from human bone marrow and transfected with chitosan-pDNA nanoparticles. The cultures transfected by lipofectamine(2000) were taken as the control. Cell viability was determined by MTT assay and transfection efficiency by flow cytometry.

RESULTS: The smallest size of complexes was obtained with 18 KD chitosan (211 nm) and the highest zeta potential was observed with 136 KD chitosan (29.61 mV). The best transfection rate (18.43%) was achieved with the 0.1% concentration of 18 KD chitosan nanoparticles versus 40.57% for commercial lipofectamine ($p < 0.01$). The MTT assay indicated an average of 95.5% cell viability for 0.1% concentration of 18 KD chitosan compared with approximately 60% of Lipofectamine(2000).

CONCLUSION: Nanoparticles produced by 18 KD chitosan at the 0.1% concentration and pDNA may be a promising gene delivery system for human marrow-derived MSCs. Although transfection efficiency of such nanoparticles is lower than that of Lipofectamine(2000), however comparatively they possess less cytotoxic effects.

KEYWORDS: chitosan; gene transfection; mesenchymal stem cells; nanoparticle; plasmid DNA

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